Efficient Differentiation of Human Pluripotent Stem Cells into Liver Cells.

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Public Summary:
Liver failure is the 12th leading cause of adult death in developed nations. Currently, end-stage liver failure can only be treated by transplanting a new liver. However, new livers are in short supply, as there is an urgent shortage of livers available from organ donors; therefore, patients with end-stage liver failure remain on the "transplant list" for extended periods of time, and sometimes never receive a life-saving liver transplant. One of our long-term goals is to produce new human liver cells in a Petri dish from embryonic stem cells, thus providing a theoretically endless supply of human liver cells for patients-in-need. This paper describes our progress thus far towards this goal, and details our laboratory's procedure to generate enriched batches of new human liver cells in a Petri dish from embryonic stem cells, with step-by-step procedures.

Scientific Abstract:
The liver detoxifies harmful substances, secretes vital proteins, and executes key metabolic activities, thus sustaining life. Consequently, liver failure-which can be caused by chronic alcohol intake, hepatitis, acute poisoning, or other insults-is a severe condition that can culminate in bleeding, jaundice, coma, and eventually death. However, approaches to treat liver failure, as well as studies of liver function and disease, have been stymied in part by the lack of a plentiful supply of human liver cells. To this end, this protocol details the efficient differentiation of human pluripotent stem cells (hPSCs) into hepatocyte-like cells, guided by a developmental roadmap that describes how liver fate is specified across six consecutive differentiation steps. By manipulating developmental signaling pathways to promote liver differentiation and to explicitly suppress the formation of unwanted cell fates, this method efficiently generates populations of human liver bud progenitors and hepatocyte-like cells by days 6 and 18 of PSC differentiation, respectively. This is achieved through the temporally-precise control of developmental signaling pathways, exerted by small molecules and growth factors in a serum-free culture medium. Differentiation in this system occurs in monolayers and yields hepatocyte-like cells that express characteristic hepatocyte enzymes and have the ability to engraft a mouse model of chronic liver failure. The ability to efficiently generate large numbers of human liver cells in vitro has ramifications for treatment of liver failure, for drug screening, and for mechanistic studies of liver disease.